

12

EUROPEAN PATENT APPLICATION

21 Application number: 86304103.4

51 Int. Cl.⁴: **C 07 C 172/00**
A 61 K 31/59

22 Date of filing: 29.05.86

30 Priority: 30.05.85 JP 117487/85

43 Date of publication of application:
17.12.86 Bulletin 86/51

84 Designated Contracting States:
AT BE CH DE FR GB IT LI LU NL SE

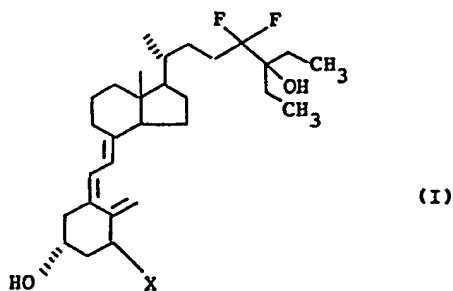
71 Applicant: **TAISHO PHARMACEUTICAL CO. LTD**
24-1 Takata 3-chome Toshima-ku
Tokyo 171(JP)

72 Inventor: **Ikekawa, Nobuo**
2-21-5, Kichijyoji Higashi-cho
Musashino-shi Tokyo(JP)

74 Representative: **Myerscough, Philip Boyd et al,**
J.A.Kemp & Co. 14, South Square Gray's Inn
London, WC1R 5EU(GB)

54 Vitamin D3 derivatives.

57 24,24-Difluoro-26,27-dimethylvitamin D₃ derivatives represented by formula:



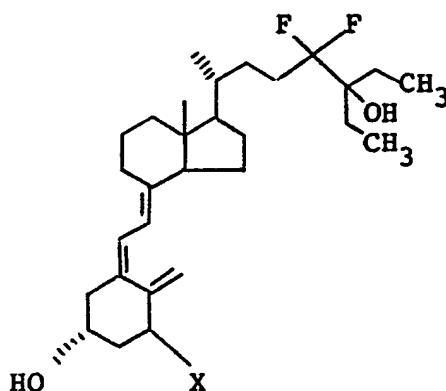
wherein

X represents a hydrogen atom or a hydroxyl group.

These compounds are prepared by irradiating 24,24-difluoro-26,27-dimethylcholesta-5,7-diene-(1 α),3 β ,25-di- or triol with ultraviolet light, and is useful for the prevention or treatment of, for example, calcium pathobolism or osteoporosis.

- 1 -
VITAMIN D₃ DERIVATIVES

This invention relates to derivatives of vitamin D₃ (i.e., cholecalciferol), and more specifically to 24,24-difluoro-26,27-dimethyl vitamin D₃ derivatives represented by the formula



wherein X represents a hydrogen atom or a hydroxyl group,

a process for production thereof, medicaments comprising the vitamin D₃ derivatives, and use of the derivatives as an active ingredient of medicaments for the prevention or treatment of diseases, such as calcium pathobolism and osteoporosis in particular.

Heretofore, the pharmacological effect of vitamin D₃ has been evaluated mainly with regard to its activity of increasing a calcium level in blood in animals with vitamin D₃ deficiency. But when it is used as a drug for improving bone symptoms in osteoporosis, renal failure and reduced function of the parathyroid gland, the blood calcium level increasing activity is not a direct activity, but is a secondary passive activity. Hypercalcemia is a side effect which appears during the treatment of the above diseases and requires the greatest caution. Frequently, it induces calcium deposition on the tissues and renal disorders, and the clinical doses of vitamin D₃ should therefore be restricted.

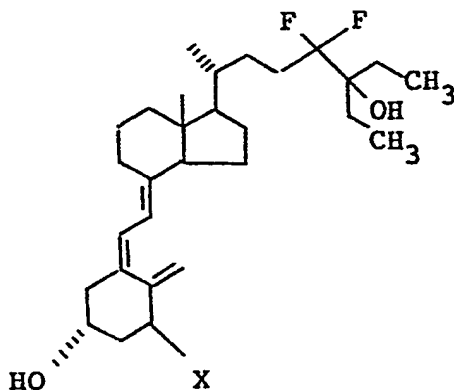
- 2 -

Accordingly, a compound having a strong activity of increasing the blood calcium level is not always useful in clinical applications. Clinical results can be anticipated more accurately by evaluating the biological activity of a vitamin D₃ derivative by using an animal disease model such as a rat osteoporosis model rather than evaluating it on the basis of the activity of increasing the blood calcium level.

When evaluated in regard to pharmacological effects and toxicity, vitamin D₃ derivatives proposed heretofore have not proved to be entirely satisfactory, and it is desired to develop new vitamin D₃ derivatives having strong bone forming activity.

The present inventor have extensively worked for vitamin D₃ derivatives having excellent pharmacological effects, particularly excellent bone forming activity, and low toxicity, and have now found that by introducing two fluorine atoms into the carbon atom at the 24-position of vitamin D₃ and a methyl group into the 26- and 27-positions respectively, novel vitamin D₃ derivatives can be obtained which in spite of their low toxicity, have a stronger bone forming activity than known 1,25-dihydroxy-26,27-dimethylvitamin D₃ and 1,25-dihydroxy-24,24-difluorovitamin D₃ in evaluations with a osteoporosis model.

Thus, according to this invention, there are provided 24,24-difluoro-26,27-dimethyl vitamin D₃ derivatives represented by the formula



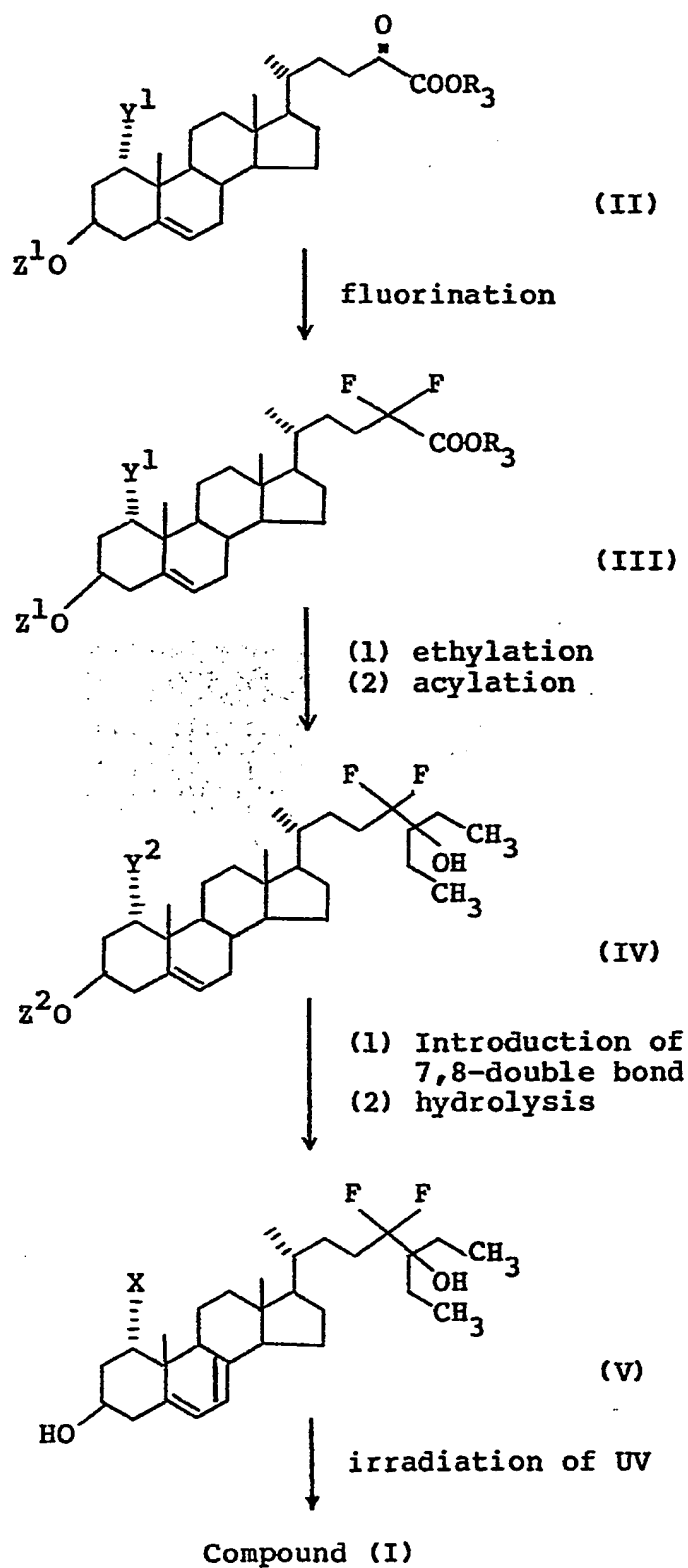
(I)

- 3 -

wherein X represents a hydrogen atom or
a hydroxyl group
as novel compounds.

The compound of formula (I) can be produced from
5 compounds of formula (II) via the following reaction route.

- 4 -

Reaction scheme A

- 5 -

In the above formulae,
 y^1 and y^2 are identical or different and
each represents a hydrogen atom or an acyloxy group;
 z^1 and z^2 are identical or different and

5 each represents an acyl group;

R represents a lower alkyl group; and
X is as defined hereinabove.

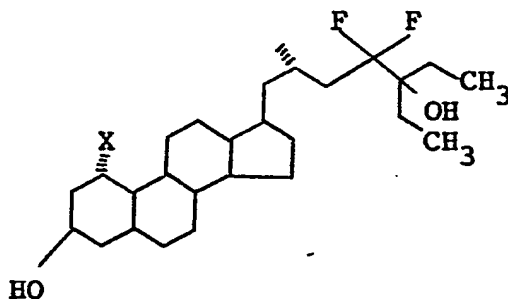
The acyl moiety of the "acyloxy group" and the
"acyl group" are, for example, lower alkanoyl groups such
10 as formyl, acetyl, propionyl and butyryl groups, and
aralkanoyl groups such as a benzoyl group. The "lower
alkyl group" includes, for example, linear or branched
alkyl groups such as methyl, ethyl, or propyl groups,
the methyl group being especially preferred.

15 The term "lower", as used in the present specification and claims, means that a group or compound qualified by this term has not more than 6, preferably not more than 4, carbon atoms.

The fluorination of the compound of formula
20 (II) is carried out by treating the compound of formula
(II) with (diethylaminosulfur) trifluoride as a fluorinating agent usually in a suitable solvent such as dichloromethane to introduce two fluorine atoms into the carbon atom at the 24-positions. Generally, the fluorination is
25 carried out at room temperature in an inert gaseous atmosphere.

The resulting compound of formula (III) is then ethylated. Usually, the ethylation is carried out by reacting the compound of formula (III) with ethyllithium
30 in a solvent such as tetrahydrofuran in an inert gaseous atmosphere. By this ethylation, the alkanoyl group at the 3-position (when y^1 at the 1-position is an alkanoyloxy group, its alkanoyl moiety as well) is split off simultaneously with the ethylation of the carbon atom at
35 the 25-position to form a compound of the following compound.

- 6 -



wherein X is as defined above. Hence, this compound is acylated in a customary manner to protect the hydroxyl group.

Thus, the compound of formula (IV) is obtained.

5 Then, a double bond is introduced between the 7- and 8-positions of this compound, and an acyl group at its 3 β -position (when an acyl group exists at the 1 α -position the acyl group as well) is split off by hydrolysis. The introduction of the double bond is effected, for example,
 10 by brominating the compound of formula (IV) with N-bromosuccinimide to introduce a bromine atom into the carbon atom at the 7-position, and thereafter dehydrobrominating the brominated compound. The reaction of the compound of formula (IV) with N-bromosuccinimide can be carried out
 15 by heating them at the refluxing temperature in an inert gaseous atmosphere in a solvent such as carbon tetrachloride.

The dehydrobromination of the 7-brominated compound may be carried out by a method known per se, for
 20 example by treating it with tetra-n-butyl ammonium fluoride at room temperature in the presence of a catalytic amount of tetra-n-butyl ammonium bromide in a solvent such as tetrahydrofuran in an atmosphere of an inert gas while shutting off light.

25 The resulting 5,7-diene compound is hydrolyzed to split off the acyl group at the 3 β -position and if

- 7 -

present, the acyl group at the 1 α -position. Hydrolysis can be carried out by a method known per se, for example by treating the 5,7-diene compound with an alcohol solution of an alkali hydroxide such as potassium hydroxide and sodium hydroxide.

The resulting compound of formula (V) can be converted into the 24,24-difluoro-26,27-dimethyl vitamin D₃ derivative of formula (I) by irradiation of ultraviolet light.

The ultraviolet light irradiation of the compound of formula (V) can be carried out by dissolving it in a suitable solvent, preferably a low-boiling solvent, and irradiating the solution with ultraviolet light. Examples of suitable solvents are hydrocarbons such as hexane, octane and benzene, ethers such as diethyl ether and tetrahydrofuran, alcohols such as methanol and ethanol, preferably lower alkanols, and mixtures thereof. The effective wavelength of ultraviolet light used for irradiation is from 200 to 360 nm. Lights of any light sources are available if they include the ultraviolet light having the wavelengths in the above range. Examples of suitable light sources are a medium-pressure mercury lamp, a high-pressure mercury lamp and laser beams. If required, unwanted lights may be cut through a filter. The time for irradiation of ultraviolet light varies with the intensity of a lamp as a light source and the scale of reaction, and is properly selected from a range of between several tens of seconds and several hours.

The irradiation is performed at temperatures in the range of usually from about -20 to about 120°C, preferably from about -10 to about 30°C, in an atmosphere of preferably an inert gas.

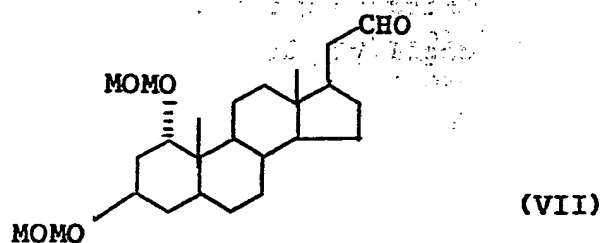
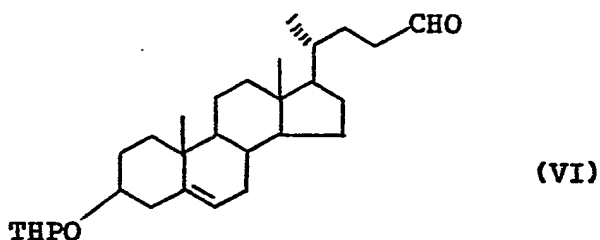
After the irradiation, the solution is kept in an inert gaseous atmosphere at temperatures from room temperature to the reflux temperature, preferably the

- 8 -

reflux temperature, for 1 to 2 hours to give a 24,24-difluoro-26,27-dimethyl vitamin D₃ derivative.

The product can be separated from the reaction mixture and purified by a method known per se, such as chromatography, extraction or recrystallization.

The compound of formula (II) used as a starting compound in the process steps shown in reaction scheme A can be synthesized by using a known compound of formula (VI) or (VII).



In the formulae, THP represents a tetrahydropyranyl group, and MOM, a methoxymethyl group.

A compound of formula (II) in which Y¹ is a hydrogen atom can be obtained by reacting the compound of formula (VI) with a tris(methylthio)methyl anion, treating the product with an acid to remove the tetrahydropyranyl group (THP) to form a diol, selectively acylating the diol with an acyl chloride in pyridine to form a 3 β -acyloxy-24-ol compound, hydrolyzing the resulting compound

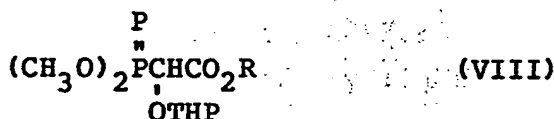
15

20

- 9 -

with mercuric chloride/mercuric oxide in an aqueous lower alkanol, particularly aqueous methanol to obtain an -hydroxy ester, and oxidizing the ester by the method of Swern et al. [J. Org. Chem., vol. 43, page 2480 (1975)].

A compound of formula (II) in which Y^1 is an acyloxy group can be obtained by subjecting the compound of formula (VII) and methylenetriphenylphosphorane to the Wittig reaction, selectively hydroborating the resulting product with 9-borabicyclo[3,3,1] nonane, oxidizing the product with hydrogen peroxide under alkaline conditions to form a 23-ol compound, oxidizing the 23-ol compound by the method of Swern et al. to form a 23-aldehyde compound, removing the methoxymethyl group (MOM) from the aldehyde compound, acylating the resulting compound, thereafter treating the product with an anion obtained from a phosphonoacylate derivative of the following formula



wherein R and THP are as defined above, and n-butyllithium to form an enol ether, and treating it with an acid.

The 24,24-difluoro-26,27-dimethyl vitamin D_3 derivatives provided by this invention have excellent bone forming activity in mammals, and is less toxic. Accordingly, the compound of this invention is useful as a medicament for the prevention and/or treatment of various diseases caused by abnormality and disorders of calcium balance and calcium absorption, such as calcium pathobolism and osteoporosis.

The excellent activity of the compound of this invention can be demonstrated by the following animal test.

- 10 -

Measurement of bone forming activity in a rat
osteoporosis model

Test animals:-

Wistar-strain female rats (28 to 32 weeks old)
5 were purchased. Eleven weeks later (39 to 43 weeks old),
the rats got ovariectomy and sciatic neurotom under
pentobarbital anesthesia. Thus, experimental osteoporosis
was induced. Ten rats per group were used.

Experimental method:-

10 From 3 to 7 days after the operation, a test
compound dissolved in Panacede 810 was orally administered
to rats over the course of 10 weeks at a dose of 0.5 ml
per day per kg of body weight four times a week (on
Monday, Tuesday, Thursday, Friday). On the day next to
15 the day of final administration of the test compound, the
femur was removed, defatted with ethanol and ether, dried
at 110°C for 16 hours, and ashed at 900°C for 16
hours. The amount of calcium in the ash was measured by
atomic absorptiometry (the amount of bone calcium). The
20 amount of inorganic phosphorus in the ash (the amount of
bone inorganic phosphorus was measured by the Fiske-
Subbarrow method [C. H. Fiske and YU. Subbarow: J. Biol.
Chem., 66, 375 (1925)]).

Shimazu double beam atomic absorptiometer
25 AA-650 was used in atomic absorptiometry, and Hitachi
500-type automatic analyzer was used in the Fiske-
Subbarrow method.

The test compounds and doses are shown in Table
1. In Table 1, the control is a vehicle for the test
30 compound. Compound A is 1 α ,25-dihydroxy-24,24-difluoro-
26,27-dimethyl vitamin D₃.

Test results:-

The results are summarized in Table 1. It is
seen that compound A of this invention showed an activity
35 of markedly increasing the amount of bone calcium and the
amount of bone inorganic phosphorus, and excellent bone

- 11 -

forming activity.

Table 1

Test compound	Dose (pmole/kg)	Amount of bone Ca * (mg)	Amount of bone inorganic P * (mg)
Control	0	120.4±3.36	62.3±1.57
Compound A	200	138.8±4.00	72.3±1.90

*: The values are the average \pm standard error.

For use as a medicament for the prevention
5 and/or treatment of diseases such as calcium pathobolism
and osteoporosis, the compound of this invention may be
administered to mammals in a dose of about 25 to about
400 ng/day, preferably about 50 to 200 ng/day. This dose
range, however, is a provisional criterion, and the
10 compound may be administered in doses outside this range
by a physician's judgement depending upon the condition,
sex, age and weight, for example, of a patient. The
administration may be effected orally or parenterally
through various routes (e.g., subcutaneous, intramuscular,
15 intravenous, intraperitoneal, and intrarectal).

The compound of this invention may be formu-
lated into a dosage form according to the route of
administration. For example, for oral administration, it
can be formulated into tablets, capsules, granules,
20 powders, syrups, elixirs, etc. For parenteral adminis-
tration, it can be formulated into injectable prepara-
tions, drops, suppositories, etc. A pharmaceutical
composition in such dosage forms may be prepared by
mixing an effective amount of the compound of this in-
25 vention with a pharmaceutically acceptable carrier or
diluent (adjuvant) and formulating the mixture into the

- 12 -

desired dosage form in a usual manner.

Illustrative of the adjuvant which may be incorporated in solid preparations such as tablets, capsules, granules and powders are binders such as tragacanth gum, acacia, corn starch or gelatin; excipients such as dicalcium phosphate; disintegrants such as corn starch, potato starch or alginic acid; lubricants such as magnesium stearate; sweetening agents such as sucrose, lactose or saccharin; and flavoring agents such as peppermint, oil of wintergreen or cherry. Various other materials may be present as coatings or in order to otherwise modify the physical form of the dosage unit. For example, tablets may be coated with shellac, sugar, or both. The syrup or elixir may contain the active compound, sucrose as a sweetening agent, methyl and propyl parabens as preservatives, a dye and a cherry or orange flavor.

Sterile compositions for injection can be formulated according to the conventional practice of pharmaceutical preparation by dissolving or suspending the active compound in a vehicle such as water for injection, a natural vegetable oil such as sesame oil, coconut oil, peanut oil or cottonseed oil, or a synthetic fatty vehicle such as ethyl oleate. Buffers, preservatives, antioxidants and the like may be incorporated as required.

The following Examples illustrate this invention in more detail.

EXAMPLE 1

(a) Synthesis of 3 β ,24 ξ -dihydroxy-24-tris(methylthio)methylchol-5-ene:-

Under an argon atmosphere at -78°C, 10.4 ml (17.7 mmoles) of a hexane solution of n-butyllithium was added to a solution of 2.4 ml (18.0 mmoles) of tris(methylthio)methane in 40 ml of tetrahydrofuran (THF for short), and the mixture was stirred for 40 minutes at the

- 13 -

same temperature. To the mixture was added dropwise 15 ml of a THF solution of 798 mg (1.8 mmoles) of 3 β -hydroxychol-5-en-24-al [compound of formula (VI)], and the mixture was stirred at -78°C for 40 minutes. The mixture was extracted with ether in a customary manner, and the resulting residue was dissolved in a mixture of 10 ml of THF and 5 ml of methanol. Four drops of 2N hydrochloric acid was added, and the mixture was stirred at room temperature for 100 minutes. The mixture was extracted with ether in a customary manner, and the resulting crude product was purified by silica gel column chromatography [31 g; eluted with hexane/ethyl acetate (10:1)] to give 651 mg of the captioned compound having a melting point of 141 to 143°C (recrystallized from methanol).

(b) Synthesis of 3 β -acetoxy-24-tris(methylthio)-methylchol-5-en-24 ξ -ol:-

The 3 β ,24 ξ -diol compound obtained in (a) above (103 mg; 0.20 mmole) was dissolved in 0.5 g of pyridine, and 15.6 microliters (0.22 mmole) of acetyl chloride was added. The mixture was stirred at room temperature for 50 minutes. The mixture was extracted with ether in a customary manner. The resulting crude product was purified by silica gel column chromatography [14 g; eluted with hexane/ethyl acetate (10:1)] to give 77 mg of the captioned acetoxy compound having a melting point of 139 to 141°C (recrystallized from methanol).

(c) Synthesis of methyl 3 β -acetoxy-24 ξ -hydroxychol-5-ene-24-carboxylate:-

A mixture of 287 mg (0.52 mmole) of the 24 ξ -ol compound obtained in (b) above, 447 mg (1.65 mmoles) of mercuric chloride and 151 mg (0.71 mmole) of mercuric oxide was stirred at room temperature for 2 hours in aqueous methanol (methanol:water=12:1). The reaction mixture was suction-filtered, and the residue was washed with dichloromethane. The filtrate was extracted with

- 14 -

ether. The organic layer was washed with a saturated aqueous solution of ammonium acetate, and worked up in a customary manner. The resulting crude product was purified by silica gel column chromatography [29 g; eluted with hexane/ethyl acetate(5:1)] to give 230 mg of the captioned methyl carboxylate having a melting point of 120 to 122°C (recrystallized from hexane).

(d) Synthesis of methyl 3 β -acetoxy-24-oxochol-3-ene-24-carboxylate:-

Dimethyl sulfoxide (0.13 ml; 0.91 mmole) was added to 4 ml of a dichloromethane solution of 0.08 ml (0.91 mmole) of oxalyl chloride at -78°C in an atmosphere of argon, and the mixture was stirred for 10 minutes under the same conditions. To the solution was added 5 ml of a dichloromethane solution of 217 mg (0.47 mmole) of the 24 ξ -ol compound obtained in (c) above, and the mixture was stirred for 15 minutes. Triethylamine (0.52 ml; 3.75 mmoles) was added, and the mixture was stirred for 5 minutes. Then, the temperature of the reaction mixture was returned to room temperature. The mixture was extracted with ether in a customary manner, and the resulting crude product was purified by silica gel column chromatography [31 g; eluted with hexane/ethyl acetate (5:1)] to give 148 mg of the captioned α -keto ester [compound of formula (II) in which Y¹ is H] having a melting point of 140 to 142°C (recrystallized from hexane/ethyl acetate).

(e) Synthesis of methyl 3-acetoxy-24,24-difluorochol-5-ene-24-carboxylate:-

(Diethylaminosulfur) trifluoride (DAST) (1.8 ml; 13 mmoles) was added at 0°C in an argon atmosphere to a solution of 848 mg (1.85 mmoles) of methyl 3 β -acetoxy-24-oxochol-5-ene-24-carboxylate, and the mixture was stirred at room temperature for 45 hours. The mixture was extracted with ethyl acetate in a customary manner, and the resulting crude product was purified by silica

- 15 -

gel column chromatography [109 g; eluted with hexane/ethyl acetate (20:1)] to give 802 mg of the captioned 24,24-difluoro compound having a melting point of 125 to 127°C (recrystallized from hexane).

- 5 (f) Synthesis of 3 β -acetoxy-24,24-difluoro-26,27-dimethylcholest-5-en-25-ol:-

An ethyl lithium solution (6.5 ml; about 8.5 mmoles) prepared from 1.9 ml (25 mmoles) of ethyl bromide and 0.75g (about 110 mmoles) of lithium was added at 10 0°C in an argon atmosphere to 7 ml of a THF solution of 80 mg (0.17 mmole) of methyl 3 β -acetoxy-24,24-difluorochol-5-ene-24-carboxylate, and the mixture was stirred for 1.5 hours under the same conditions. The mixture was ex-tracted with ethyl acetate in a customary manner, and the 15 resulting residue was stirred at room temperature for 12 hours with 0.5 ml of acetic anhydride in 0.5 ml of pyri-dine, and then extracted with ethyl acetate in a customary manner by silica gel column chromatography [4.6g; hexane/ethyl acetate (20:1)] to give 62 mg of the captioned 20 26,27-dimethyl compound having a melting point of 127 to 128°C (recrystallized from methanol).

- (g) Synthesis of 24,24-difluoro-3 β ,25-di hydroxy-26,27-dimethylcholesta-5,7-diene:-

A mixture of 25.8 mg (0.051 mmole) of 3 β - 25 acetoxy-24,24-difluoro-26,27-dimethylcholest-5-ene-25-ol and 12.7 mg (0.017 mmole) of N-bromosuccimide was heated under reflux for 25 minutes in 3.5 ml of carbon tetra-chloride in an atmosphere of argon. The reaction mixture was cooled to 0°C. The precipitate formed was separated 30 by filtration, and the solvent was evaporated from the filtrate. The resulting residue was dissolved in 4 ml of THF, and a catalytic amount of tetra-n-butyl ammonium bromide was added. The mixture was stirred for 50 minutes in an atmosphere of argon while shutting off light. 35 Then, 0.16 ml (0.16 mmole) of a 1M THF solution of tetra-

- 16 -

n-butyl ammonium fluoride was added and the mixture was stirred for 30 minutes under the same conditions. The mixture was extracted with ethyl acetate in a customary manner, and the resulting crude product was purified by
5 thin-layer silica gel chromatography (developing solvent: hexane/ethyl acetate=10:1; developed three times) to give 11.3 mg of the acetate. It was dissolved in 4 ml of THF, and 1 ml of a 5% methanol solution of potassium hydroxide
10 for 30 minutes in an atmosphere of argon while shutting off light. The mixture was extracted with ethyl acetate in a customary manner, and the resulting crude product was purified by thin-layer silica gel chromatography (developing solvent: hexane/ethyl acetate=4:1; developed
15 four times) to give 9.3 mg of the captioned 5,7-diene compound.

UV $\lambda_{\text{max}}^{\text{EtOH}}$: 294, 282, 272 nm.

(h) Synthesis of 24,24-difluoro-25-hydroxy-26,27-dimethyl vitamin D₃:-

20 9.3 mg of 24,24-difluoro-3 β ,25-dihydroxy-26,27-dimethylcholesta-5,7-diene was dissolved in a mixture of 90 ml of benzene and 40 ml of ethanol, and in an argon stream at 0°C, the solution was irradiated with ultra-violet light for 7 minutes using a medium-pressure mercury
25 lamp through a Vycor filter. Subsequently, the reaction mixture was heated under reflux for 1 hour in an atmosphere of argon. The solvent was evaporated under reduced pressure, and the resulting crude product was purified by thin-layer silica gel chromatography (developing solvent:
30 benzene/ethyl acetate=10:1; developed four times) to give 2.6 mg of the captioned vitamin D₃ derivative [the compound of formula (I) in which X is H].

UV $\lambda_{\text{max}}^{\text{EtOH}}$: 265 nm, $\lambda_{\text{min}}^{\text{EtOH}}$: 228 nm.

- 17 -

EI-MS (70eV) m/z:

464 (M, 32, 3), 446 (9, 1), 444 (2, 8),
431 (23, 3), 271 (17, 2), 253 (21, 6),
175 (6, 9), 173 (10, 1), 155 (11, 2),
5 136 (76, 5), 118 (base peak), 87 (37, 1),
6.9 (4, 5).

EXAMPLE 2

(a) Synthesis of $1\alpha,3\beta$ -dimethoxymethoxy-22-norchol-5,22-diene:-

10 In an argon stream at 0°C, 1.9 ml (3.23
mmoles) of a 1.7M hexane solution of n-butyllithium was
added to 12 ml of a THF suspension of 1.4 g (3.45 mmoles)
of methyltriphenyl phosphonium iodide, and the mixture
was stirred for 50 minutes. To the mixture was added
15 dropwise at 0°C 8 ml of a THF solution of 1.0 g (2.32
mmoles) of the compound (VII), and the mixture was stirred
at room temperature for 30 minutes. The mixture was
extracted with ether in a customary manner, and the
resulting product was purified by silica gel column
20 chromatography [57 g; eluted with hexane/ethyl acetate
(5:1)] to give 0.97 g of the captioned diene compound as
an oil.

(b) Synthesis of $1\alpha,3\beta$ -dimethoxymethoxy-22-norchol-5-en-23-ol:-

25 To 5 ml of a THF solution of 393 mg (0.91
mmole) of the $1\alpha,3\beta$ -dimethoxymethoxy-22-norchol-5,22-
diene obtained in (a) above was added dropwise 3.8 ml
(1.8 mmoles) of a 0.5M THF solution of 9-borabicyclo-
[3,3,1]-nonane in an atmosphere of argon at 0°C. After
30 the addition, the reaction solution was stirred at room
temperature for 45 hours. To the solution was added 0.4
ml of a 3N aqueous solution of sodium hydroxide, and
subsequently, 0.4 ml of a 30% aqueous solution of hydrogen
peroxide at 0°C. The mixture was stirred at the same
35 temperature for 35 minutes. The mixture was extracted
with ether in a customary manner. The resulting crude

- 18 -

product was purified by silica gel column chromatography [85 g; eluted with hexane/ethyl acetate (3:1)] to give 252 mg of the captioned 23-ol compound having a melting point of 107 to 109°C (recrystallized from hexane).

5 (c) Synthesis of 1 α ,3 β -dimethoxymethoxy-22-norchol-5-en-23-al:-

Dimethyl sulfoxide (0.41 ml; 5.79 mmoles) was added to 10 ml of a dichloromethane solution of 0.28 ml (2.88 mmoles) of oxalyl chloride at -78°C in an atmosphere of argon, and the mixture was stirred for 10 minutes under the same conditions. To the mixture was added 6 ml of a dichloromethane solution of 850 mg (1.44 mmoles) of 1 α ,3 β -dimethoxymethoxy-22-norchol-5-en-23-ol, and the mixture was stirred for 15 minutes. Triethylamine (1.6 ml; 11.5 mmoles) was added, and the mixture was stirred for 5 minutes. Then, the temperature of the reaction mixture was returned to room temperature. The reaction mixture was extracted with ether in a customary manner. The resulting crude product was purified by silica gel column chromatography [40 g; hexane/ethyl acetate (4:1)] to give 579 mg of the captioned aldehyde as an oil.

(d) Synthesis of 1 α ,3 β -diacetoxy-22-norchol-5-en-23-al:-

6N hydrochloric acid (2.4 ml) was added to 8 ml of a THF solution of 579 mg (1.3 mmoles) of 1 α ,3 β -dimethoxymethoxy-22-norchol-5-en-23-al, and the mixture was stirred at 60°C for 60 minutes. The mixture was extracted with ethyl acetate in a customary manner. The resulting residue was dissolved in 3 ml of pyridine, and stirred with 1 ml of acetic anhydride at room temperature for 20 hours. The mixture was extracted with ethyl acetate in a customary manner, and the resulting crude product was purified by silica gel column chromatography [37; eluted with hexane/ethyl acetate (4:1)] to give 258 mg of the captioned diacetate compound having a melting point of 135 to 137°C (recrystallized from hexane/ethyl acetate).

- 19 -

(e) Synthesis of methyl $1\alpha,3\beta$ -diacetoxy-24-oxochol-5-en-24-carboxylate:-

To 1.5 ml of a THF solution of 0.13 ml (1.12 mmoles) of diisopropylamine was added 0.6 ml (1.02 mmoles) of an n-butyl lithium solution at -78°C in an atmosphere of argon. The mixture was stirred for 30 minutes to prepare an LDA solution. To the solution was added 4 ml of a THF solution of 310 mg (1.10 mmoles) of the phosphonoacetate [the compound of formula (VIII) in which R is methyl], and the mixture was stirred at -78°C for 30 minutes. Then, 4 ml of a THF solution of 377 mg (0.85 mmole) of the $1\alpha,3\beta$ -diacetoxy-22-norchol-5-en-23-al obtained in (c) above, and the mixture was stirred at room temperature for 2.5 hours. The mixture was extracted with ether in a customary manner. The resulting crude enol ether was dissolved in a mixture of 4 ml of THF and 4 ml of methanol, and 20 mg of p-toluenesulfonic acid was added. The mixture was stirred at room temperature for 40 minutes. The mixture was extracted with ether in a customary manner, and the resulting crude product was purified by silica gel column chromatography [40 g; eluted with hexane/ethyl acetate (7:1)] to give 225 mg of the captioned α -keto ester (I) [the compound of formula (II) in which Y^1 is acetoxy] as an oil.

25 (f) Synthesis of methyl $1\alpha,3\beta$ -diacetoxy-24,24-difluorochol-5-en-24-carboxylate:-

By the same operation as in Example 1, (e), the captioned difluoro compound (as an oil) was obtained from the methyl $1\alpha,3\beta$ -diacetoxy-24-oxochol-5-ene-24-carboxylate obtained in (e) above.

(g) Synthesis of $1\alpha,3\beta$ -diacetoxy-24,24-difluoro-26,27-dimethylcholest-5-en-25-ol:-

By the same operation as in Example 1, (f), the captioned dimethyl compound (as an oil) was obtained from methyl $1\alpha,3\beta$ -diacetoxy-24,24-difluoro-5-ene-25-carboxylate obtained in (f) above.

- 20 -

(h) Synthesis of 24,24-difluoro-26,27-dimethyl-1 α ,3 β ,25-trihydroxycholesta-5,7-diene:-

By the same operation as in Example 1, (g), the captioned 5,7-diene compound was obtained from 1 α ,3 β -diacetoxy-24,24-difluoro-26,27-dimethylcholest-5-en-25-ol obtained in (g) above.

UV $\lambda_{\text{max}}^{\text{EtOH}}$: 294, 282, 271, 262 nm.

(i) Synthesis of 1 α ,25-dihydroxy-24,24-difluoro-26,27-dimethyl vitamin D₃:-

The 24,24-difluoro-26,27-dimethyl-1 α ,3 β ,25-trihydroxycholesta-5,7-diene (1.0 mg) was dissolved in a mixture of 90 ml of benzene and 40 ml of ethyl acetate, and the solution was irradiated for 3 minutes with ultra-violet light in an argon atmosphere at 0°C using a medium-pressure mercury lamp. The reaction mixture was heated under reflux for 1 hour in an argon atmosphere. The solvent was evaporated under reduced pressure, and the resulting crude product was purified by thin-layer silica gel chromatography (developing solvent: benzene/ethyl acetate=1:1; developed three times) to give 202 micrograms of the captioned vitamin D₃ derivative [the compound of formula (I) in which X is OH].

UV $\lambda_{\text{max}}^{\text{EtOH}}$: 265 nm, $\lambda_{\text{min}}^{\text{EtOH}}$: 228 nm.

EI-MS (70 eV) m/z:

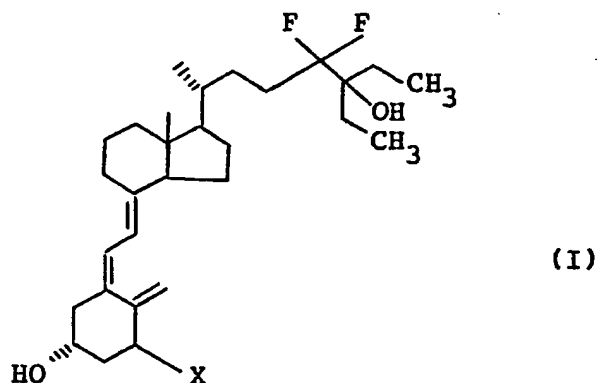
480 (M, 8, 1), 462 (75, 2), 444 (37, 0),
429 (3, 6), 287 (5, 1), 269 (28, 7, 10, 7),
251 (17, 1), 236 (4, 1), 155 (31, 0),
152 (22, 0).

134 (base peak), 116 (4,5), 87 (38, 5), 69 (4,5)

- 21 -

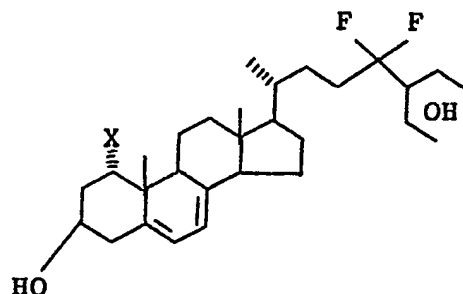
What is claimed is:

1. A 24,24-Difluoro-26,27-dimethyl vitamin D₃ derivative represented by the formula:



wherein X represents a hydrogen atom or a hydroxyl group.

2. A process for producing the 24,24-difluoro-26,27-dimethyl vitamin D₃ derivative of claim 1, which comprises irradiating a compound of the formula



wherein X represents a hydrogen atom or a hydroxyl group,

with ultraviolet light.

3. A process for producing 1,25-dihydroxy-26,27-dimethylcholecalciferol of claim 1, which comprises irradiating 26,27-dimethylcholesta-5,7-diene-1,3,25-triol with ultraviolet light.

4. The compound of claim 1 as a pharmaceutically active substance.

- 22 -

5. The compound of claim 1 as a pharmaceutically active substance for the prevention or treatment of calcium pathobolism or osteoporosis.

6. A pharmaceutical composition for the prevention or treatment of calcium pathobolism or osteoporosis, which comprises an amount, effective for the prevention or treatment of said diseases, of 24,24-difluoro-26,27-dimethylvitamin D₃ derivative of claim 1, and a pharmaceutically acceptable diluent or carrier.

7. A medicament containing 24,24-difluoro-26,27-dimethylvitamin D₃ drivative of claim 1.

8. A method for the prevention or treatment of calcium pathobolism or osteoporosis, which comprises administering an effective amount of 24,24-difluoro-26,27-dimethylvitamin D₃ derivative of claim 1 to a mammal.

9. Use of the compound of claim 1 for the prevention or treatment of calcium pathobolism or osteoporosis.



European Patent
Office

EUROPEAN SEARCH REPORT

0205285

Application number

EP 86 30 4103

DOCUMENTS CONSIDERED TO BE RELEVANT			
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int. Cl.4)
A	US-A-4 201 881 (HECTOR F. DELUCA) * Whole document *	1-9	C 07 C 172/00 A 61 K 31/59
A	US-A-4 196 133 (HECTOR F. DELUCA) * Whole document *	1-9	
P,A	EP-A-0 171 990 (TAISHO) * Claims *	1-9	
			TECHNICAL FIELDS SEARCHED (Int. Cl.4)
			C 07 C 172/00
The present search report has been drawn up for all claims			
Place of search THE HAGUE		Date of completion of the search 27-08-1986	Examiner HENRY J.C.
<p>CATEGORY OF CITED DOCUMENTS</p> <p>X : particularly relevant if taken alone Y : particularly relevant if combined with another document of the same category A : technological background O : non-written disclosure P : intermediate document</p> <p>T : theory or principle underlying the invention E : earlier patent document, but published on, or after the filing date D : document cited in the application L : document cited for other reasons & : member of the same patent family, corresponding document</p>			

**This Page is Inserted by IFW Indexing and Scanning
Operations and is not part of the Official Record**

BEST AVAILABLE IMAGES

Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images include but are not limited to the items checked:

- ☐ BLACK BORDERS
- ☐ IMAGE CUT OFF AT TOP, BOTTOM OR SIDES
- ☐ FADED TEXT OR DRAWING
- ☒ BLURRED OR ILLEGIBLE TEXT OR DRAWING
- ☐ SKEWED/SLANTED IMAGES
- ☐ COLOR OR BLACK AND WHITE PHOTOGRAPHS
- ☐ GRAY SCALE DOCUMENTS
- ☐ LINES OR MARKS ON ORIGINAL DOCUMENT
- ☐ REFERENCE(S) OR EXHIBIT(S) SUBMITTED ARE POOR QUALITY
- ☐ OTHER: _____

IMAGES ARE BEST AVAILABLE COPY.

As rescanning these documents will not correct the image problems checked, please do not report these problems to the IFW Image Problem Mailbox.